Passive and Active Maternal Smoking as Measured by Serum Cotinine: The Effect on Birthweight

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ABSTRACT

To determine how maternal exposure to environmental tobacco smoke affects birthweight, maternal sera obtained from 3529 pregnant women around 27 weeks gestation were analyzed for cotinine, a metabolite of nicotine. Based on cotinine levels, nonsmokers were divided into those exposed to environmental tobacco smoke (2–10 ng/mL) and those unexposed (<2 ng/mL), and smokers were divided into tertiles. Compared with unexposed nonsmokers' infants, infants of exposed nonsmokers averaged 45 g less ($P = .28$) after adjustment for confounders, and smokers' infants averaged 78, 191, and 233 g less for the first, second, and third cotinine tertiles, respectively. Birthweight decreased 1 g for every nanogram per milliliter of cotinine increase ($P < .001$). (*Am J Public Health.* 1995;85:395–398)

Introduction

Maternal cigarette smoking is the single most preventable cause of low birthweight.[1–3] Pregnant women exposed to others' cigarette smoke may also be more likely to deliver a low-birthweight infant.[4,5]

A number of studies[6–13] have investigated the effects of environmental tobacco smoke exposure using the woman's self-report. Yet self-report may lead to imprecise exposure assessment.[14] Only one previous study has used cotinine, a biomarker of nicotine, to provide a more accurate assessment of the relationship between environmental tobacco smoke exposure and birthweight. Haddow et al.[15] found that the mean birthweight of infants whose nonsmoking mothers had second trimester serum cotinine levels of 1.1 to 9.9 ng/mL was 108 g lower (95% confidence interval [CI] = −173, −35) than that of infants of unexposed women. However, the authors were unable to control for gestational age, a major determinant of birthweight.

The present study assesses the relationship between birthweight and environmental tobacco smoke using serum cotinine levels and controlling for potential confounders and gestational age.

Methods

Participants

The study population was composed of pregnant women who participated in the Child Health and Development Studies in Oakland, Calif, between 1964 and 1967.[16] Women who delivered a singleton live birth between 20 and 44 weeks of gestation and for whom smoking status, birthweight, and gestational age were known were included in the sample (n = 3896).

Smoking status was ascertained at an interview during early pregnancy. Most women (71%) also completed a second interview later in their pregnancy in which they were asked about changes in their smoking status. For the present study, women who quit smoking sometime during pregnancy (n = 318) were excluded. Otherwise, women who were current smokers each time they were interviewed were classified as smokers, while those who had never smoked or who had quit before pregnancy were classified as nonsmokers. This resulted in a total of 1286 smokers and 2292 nonsmokers.

The most vulnerable period for the effects of cigarette smoke appears to be in the third trimester, when the fetal growth spurt occurs.[17,18] Consequently, information on the number of cigarettes smoked was obtained from the second interview, if available, and cotinine levels were analyzed for the sera collected at around the same time (averaging 28 and 27 weeks after last menstrual period, respectively).

Covariates

Other information obtained from the first interview includes maternal age, height, prepregnancy weight, race, date of last menstrual period, alcohol and coffee consumption, education, parity, employment status, and husband's smoking status. Body mass index was determined by dividing prepregnancy weight by height squared (kilograms/square meters). Pregnancy weight gain was obtained by subtracting prepregnancy weight from weight at last prenatal visit.

Information obtained from the infant's medical chart included sex and birthweight (ounces converted to grams). Preterm delivery was defined as delivery before 37 weeks gestation, based on last menstrual period. Infants weighing less than 2500 g were considered low birthweight.

Cotinine Analysis

Cotinine is a by-product of nicotine metabolism[19]; approximately 70% of nicotine is converted into serum cotinine.[20,21]

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TABLE 1—Distribution of Sample Characteristics among Smoking Cotinine Exposure Groups as Indicated by Serum Cotinine Levels

	% Nonsmokers, 0.0–1.9 ng/mL Cotinine (n = 2129)[a]	% Nonsmokers Exposed to Environmental Tobacco Smoke, 2–10 ng/mL Cotinine (n = 114)[a]	% Smokers (n = 1286)[a]
Age, y	(n = 2128)	(n = 114)	(n = 1285)
<24	29.2	35.1	35.1
24–29	39.7	46.5	38.7
>29	31.1	18.4	26.2
Race	(n = 2129)	(n = 114)	(n = 1286)
White	59.3	53.5	65.4
African American	27.0	43.0	28.5
Other	13.7	3.5	6.1
Mother's education	(n = 2127)	(n = 114)	(n = 1286)
<12 y	11.7	9.7	23.8
High school graduate	25.4	29.8	31.7
>12 y	62.9	60.5	44.5
Mother's employment	(n = 2120)	(n = 114)	(n = 1282)
No	55.5	36.8	50.9
Yes	44.5	63.2	49.1
Parity	(n = 2129)	(n = 114)	(n = 1286)
Primiparous	35.6	40.3	31.0
Low multiparous (1–2 births)	46.5	43.0	46.4
High multiparous (≥3 births)	17.9	16.7	22.6
Body mass index, kg/m^2	(n = 2060)	(n = 114)	(n = 1252)
<18.9	11.7	14.0	14.9
18.9–25.8	74.7	70.2	72.8
>25.8	13.5	15.8	12.4
Mother's alcohol consumption	(n = 2111)	(n = 114)	(n = 1277)
None	77.6	77.2	64.1
Any	22.4	22.8	35.9
Mother's coffee consumption	(n = 2120)	(n = 114)	(n = 1279)
None	46.1	51.8	28.8
1–2 cups/day	36.0	36.0	28.9
3+ cups/day	17.9	12.3	42.3
Husband's current smoking status	(n = 1703)	(n = 92)	(n = 1031)
None	49.6	23.9	17.2
1–9 cigarettes/day	11.1	17.4	10.5
10–19 cigarettes/day	11.9	18.5	19.9
20–29 cigarettes/day	20.1	32.6	36.2
30+ cigarettes/day	7.3	7.6	16.3
Infant sex	(n = 2129)	(n = 114)	(n = 1286)
Male	50.3	47.4	51.2
Female	49.7	52.6	48.8

[a]Numbers may differ for each characteristic because of missing values.

Cotinine levels were determined at the American Health Foundation by the radioimmunoassay method of Haley et al.[22] after modification of a technique by Langone et al.[23] The detection limit was 2 ng/mL with an 11% coefficient of variation. Details of the cotinine analysis can be found elsewhere.[24]

Of all women who classified themselves as nonsmokers at both interviews, 49 had cotinine levels above 10 ng/mL. These women were excluded from most analyses. Nonsmokers with cotinine levels between 2 and 10 ng/mL were classified as having been exposed to environmental tobacco smoke (n = 114). Smokers were divided into tertiles based on their serum cotinine concentrations: 0 to 78 ng/mL, 79 to 165 ng/mL, and 166 to 569 ng/mL cotinine. Smokers with higher cotinine levels reported smoking more cigarettes per day.

Data Analysis

Chi-square tests and analyses of variance were used to assess the relationship between covariates and smoking categories. Mean birthweight and gestational age were compared across smoking categories with *F* tests; frequency of low birthweight and preterm deliveries were analyzed with chi-square tests for trend across cotinine groups. Potential confounders associated with birthweight were included in the multivariate linear regression models. Two types of regression models were performed. First, subjects were categorized (as described) into nonsmokers, passive smokers, and smokers (based on tertiles), and the differences in mean birthweight between these groups were assessed. These analyses were compared with those of Haddow et al.[15] Second, cotinine was measured as a continuous variable, and its effect on birthweight was examined in those subjects who had detectable cotinine levels (>2 ng/mL).

Models that included interactions between cotinine categories and covariates were tested for significance ($P < .20$); as no interactions were significant, they were excluded. A quadratic term for gestational age was included in the final analysis to improve model fit ($P < .01$). Gestational age was centered by subtracting each value from the mean and dividing the result by 100. SAS software, Version 6.04, was used.[25]

Results

Table 1 shows that women exposed to environmental tobacco smoke were more likely to work and to have husbands who smoked than were other nonsmokers; they were also slightly younger and more likely to be African American. Infants of women exposed to environmental tobacco smoke were similar on all measures of birthweight and gestational age to infants of smokers in the lowest cotinine tertile and to infants of nonsmokers (Table 2). Infants of environmental tobacco–exposed women had unadjusted birthweights that averaged 43 g less than infants of nonsmokers, but this was not significant.

In fact, shortened gestation was evident only in infants of smokers in the third (highest) cotinine tertile, while rates of low birthweight were highest for smokers in the second and third tertiles. Multivariate analyses were not conducted for low birthweight and preterm births because numbers in the environmental tobacco smoke–exposed group were small.

Table 3 presents the association between serum cotinine and birthweight, adjusting for potential confounders. Nonsmokers exposed to environmental tobacco smoke had infants that were, on

TABLE 2—Unadjusted Birthweight and Gestational Age Measurements for Infants of Women in Differing Smoking Exposure Groups as Defined by Serum Cotinine Levels

	Nonsmokers, 0.0–1.9 ng/mL Cotinine (n = 2129)	Nonsmokers Exposed to Environmental Tobacco Smoke, 2.0–10.0 ng/mL Cotinine (n = 114)	Smokers in 1st Tertile,[a] 0–78 ng/mL Cotinine (n = 425)	Smokers in 2nd Tertile,[a] 79–165 ng/mL Cotinine (n = 433)	Smokers in 3rd Tertile,[a] ≥166 ng/mL Cotinine (n = 428)	Statistic[b] *P*
Mean birthweight, g (SD)	3369.1 (518.9)	3326.3 (497.8)	3326.3 (529.4)	3185.7 (473.3)	3073.6 (527.7)	.001
Mean gestational age, d (SD)	279.6 (15.1)	279.6 (12.6)	280.1 (16.0)	279.3 (15.1)	276.2 (17.1)	.001
Low birthweight						
<2500 g, % (n)	3.9 (83)	5.3 (6)	4.5 (19)	6.2 (27)	12.9 (55)	
RR (95% CI)	. . .	1.35 (0.60, 3.03)	1.15 (0.70, 1.87)	1.60 (1.05, 2.44)	3.30 (2.38, 4.56)	.001
Preterm birth						
≤37 weeks, % (n)	6.9 (146)	7.0 (8)	6.8 (29)	7.2 (31)	10.1 (43)	
RR (95% CI)	. . .	1.02 (0.51, 2.03)	1.00 (0.68, 1.46)	1.04 (0.72, 1.52)	1.47 (1.06, 2.03)	.08

Note: RR = relative risk; CI = confidence interval.
[a]Smokers with higher cotinine levels reported smoking more cigarettes per day, resulting in 38%, 77%, and 89% of the women reporting smoking 10 or more cigarettes per day for the low, middle, and highest tertiles, respectively.
[b]χ^2 test for trend was used for categorical variables; *F* test was used for continuous variables.

average, 45 g lighter than those of other nonsmokers; however, this difference was not statistically significant. Birthweights of infants of smokers in the lowest cotinine tertile were not significantly different from those of infants of smoke-exposed nonsmokers but were different from birthweights of infants of unexposed nonsmokers.

Including sex or alcohol or coffee consumption did not improve the model, so these variables were dropped. In addition, the results did not appreciably change when we included women who reportedly were nonsmokers but had cotinine levels above 10 ng/mL (n = 49) or when we excluded women who reported smoking but had cotinine levels of not more than 10 ng/mL (n = 107) (data not shown).

The multivariate analysis of cotinine as a continuous variable was conducted on all subjects with cotinine levels of at least 2 ng/mL and included the same covariates as shown in Table 3. Results show that for each nanogram per milliliter increase in serum cotinine, birthweight decreased an average of 1.0 g (95% CI = −1.14, −0.79).

TABLE 3—Multiple Regression Model[a] for the Effects of Smoking Status/Serum Cotinine Levels on Birthweight

Independent Variable	Parameter Estimate, g	95% Confidence Interval
Environmental tobacco smoke–exposed (vs unexposed nonsmoker): 2–10 ng/mL cotinine	−44.8	−125.6, 36.0
Smoker (vs unexposed nonsmoker)		
0–78 ng/mL cotinine	−77.7	−123.2, −32.2
79–165 ng/mL cotinine	−191.0	−236.9, −145.1
≥166 ng/mL cotinine	−232.7	−279.3, −186.0
Race (vs White)		
African American	−161.6	−196.2, −127.1
Other	−169.7	−218.5, −121.0
Age (vs 24–29 y)		
<24 y	−16.9	−53.2, 19.5
>29 y	26.4	−11.2, 63.9
Parity (vs primiparous)		
1–2 births	104.1	70.2, 138.1
≥3 births	150.5	101.0, 200.0
Education (vs high school graduate)		
<12 y	−0.7	−46.9, 45.5
>12 y	37.4	2.7, 72.2
Weight gain, kg	4.8	4.2, 5.4
Body mass index (vs 18.9–25.8)		
<18.9 kg/m^2	−81.2	−124.9, −37.5
>25.8 kg/m^2	171.0	125.8, 216.2
Gestational age (per 100 days)	1104.0	978.8, 1229.3
Gestational age^2 (per 100 days)	−882.3	−1123.1, −641.4

[a]n = 3397; adjusted R^2 = 0.34.

Discussion

The results of this study suggest that, after adjustment for gestational age, nonsmoking women exposed to environmental tobacco smoke have babies that weigh slightly less (45 g) than infants of other nonsmoking women. The direction of our observed effect, although not significant, is similar to the 108-g effect of environmental tobacco smoke reported by Haddow et al.[15] The results of our study are also consistent with the moderate but often nonsignificant effects reported in studies that used questionnaire information to estimate environmental tobacco smoke

exposure.[7–13] These studies have shown a 10- to 100-g decrease in birthweight among infants of exposed women. Additionally, regardless of whether cotinine was analyzed continuously or categorically, a dose-response of decreasing birthweight with increasing serum cotinine levels was observed.

In addition to using a biomarker of nicotine exposure, our study has a number of strengths over previous studies. We were able to examine the potentially confounding effects of alcohol consumption, gestational age, and weight gain. Another strength is that, because the pregnancies occurred approximately 30 years ago, when women were less aware of the hazards of cigarette smoke than they are now, subjects may have reported their smoking habits more accurately.

The age of this study is also one of its limitations. Cotinine analysis was not performed until more than 25 years after the sera were collected. However, there is little evidence to suggest that cotinine does not remain stable over time.[22,26] Another limitation of this study is that the lower limit of detection for cotinine (2 ng/mL) was rather high. Nonsmokers with cotinine levels between 0.5 ng/mL (Haddow et al.'s[15] detection limit) and 2 ng/mL would have been misclassified as nonexposed in our study and thus could have biased our results toward the null. However, given that Haddow et al.[15] reported that birthweights were similar for infants of mothers with cotinine levels between 0.5 and 1.0 ng/mL and for infants of mothers with cotinine levels below 0.5 ng/mL, our higher limit of detection is unlikely to have affected our results.

The results of our study are biologically plausible, given that two of the major components of both mainstream and environmental tobacco smoke are carbon monoxide and nicotine, agents with known fetotoxic effects.[27] Carbon monoxide and nicotine levels, however, are lower in environmental tobacco smoke than they are in mainstream smoke.[28]

In summary, our study demonstrates a statistically nonsignificant decrement in birthweight adjusted for gestational age in infants whose mothers were exposed to environmental tobacco smoke. Although the magnitude of change is small and not of clear clinical significance, even a slight shift of a population's birthweight distribution could result in more low-birthweight infants, a group at greater risk of perinatal mortality. □

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